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Novel Application of Laboratory Instrumentation Characterizes Mass Settling Dynamics of Oil-Mineral Aggregates (OMAs) and Oil-Mineral-Microbial Interactions

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ABSTRACT

It is reasonable to assume that microbes played an important role in determining the eventual fate of oil spilled during the 2010 *Deepwater Horizon* disaster, given that microbial activities in the Gulf of Mexico are significant and diverse. However, critical gaps exist in our knowledge of how microbes influence the biodegradation and accumulation of petroleum in the water column and in marine sediments of the deep ocean and the shelf. Ultimately, this limited understanding impedes the ability to forecast the fate of future oil spills, specifically the capacity of numerical models to simulate the transport and fate of petroleum under a variety of conditions and regimes.

By synthesizing recent model developments and results from field- and laboratory-based microbial studies, the Consortium for Simulation of Oil-Microbial Interactions in the Ocean (CSOMIO) investigates (a) how microbial biodegradation influences accumulation of petroleum in the water column and in marine sediments and (b) how biodegradation can be influenced by environmental conditions and impact forecasts of potential future oil spills.

Keywords:

Laboratory Flocculation Experiments

Critical to oil-mineral-microbial interactions is a process whereby cohesive sediment particles do not behave as individual, dispersed particles but instead tend to stick together. This process is known as flocculation, and the resultant floc sizes and settling velocity are much greater than those of the individual constituent particles, but their overall floc effective density is less (e.g., Dyer & Manning, 1999; Manning & Dyer, 1999). When oil droplets are contained by flocs of cohesive sediment and/or

marine snows, oil sedimentation can occur and provide an unexpected pathway in the oil budget calculation (Daly et al., 2016; Muschenheim & Lee, 2002; Passow & Ziervogel, 2016). A novel high-resolution floc video instrument originally designed to determine the spectral characteristics of flocculating cohesive sediments has, for the first time, been applied to study floc size distribution and settling dynamics of oil-mineral aggregates (OMAs). The results of this study inform the development of efficient and accurate algorithms for simulating the formation and settling of these flocs.

As part of the Consortium for Simulation of Oil-Microbial Interactions in the Ocean (CSOMIO), a series of laboratory flocculation experiments with seawater, crude oil, and cohesive sediment mixtures (mineral clay and artificial extracellular polymeric substances) have been conducted at the Center for Applied Coastal Research, University of Delaware, using the LabSFLOC-2 (the second generation of the LabSFLOC [Laboratory Spectral Flocculation Characteristics instrument; Manning, 2015], developed by Manning, 2006). In these experiments, the LabSFLOC-2 instrument, a digital video microscope and

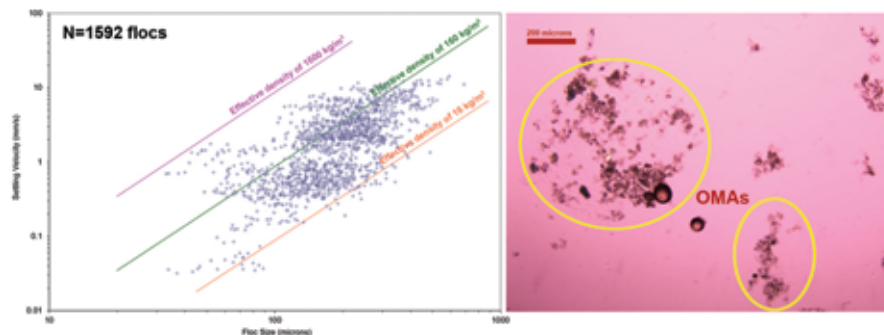
processing package, makes it possible to obtain high-quality floc population data (e.g., individual floc size, settling velocity, density, mass), as well as supplementary individual floc information including floc porosity, floc mass, fractal dimension, floc shape, and mass settling flux. Manning et al. (2010) and Manning et al. (2017) provide further details of the floc acquisition procedures and postprocessing computations, respectively. LabSFLOC-2 provides data for many important aspects of flocculation. These floc data are necessary to comprehensively assess and characterize oil-mineral-microbial settling dynamics and to improve the parameterization (Manning & Dyer, 2007; Soulsby et al., 2013) and calibration (Baugh & Manning, 2007) of numerical models. Additionally, the digital microscope images help us better understand the visible floc structure of OMAs.

Laboratory Experiments Utilizing the LabSFLOC-2 Instrument

Mass settling dynamics of oil-mineral flocs are observed using the LabSFLOC-2 system (Figure 1), which measures an entire floc population for each sample being assessed. LabSFLOC-2 utilizes a low intrusive 2.0-MP Grasshopper monochrome digital video camera to optically observe individual flocs (e.g., Manning & Dyer, 2002) as they settle in a 350 mm high \times 100 mm square Perspex settling column. The video camera, positioned nominally 75 mm above the base of the column, views all particles in the center of the column that pass within a 1-mm depth of field, 45 mm from the Sill TZM 1560 high-magnification (nominal 5- μ m pixel resolution) telecentric (maximum

FIGURE 1

The LabSFLOC-2 setup on the desk beside the stir jar system for real-time samplings (photo provided by Prof. A. J. Manning).



pixel distortion of 0.6%), 0.66 (1:1.5) magnification, F4, macro lens.

A suspension containing oil-mineral-microbial flocs is initially introduced to the LabSFLOC-2 column, while a suspension is extracted from the jar fluid using a specially modified Serological TD-EX 20°C 50-ml maximum-capacity sterile pipette. This process has proved to be minimally intrusive for flocs, relying only upon settling due to gravity and thus avoiding the need for additional fluid or turbulence transfer. The LabSFLOC-2 instrumentation is located close and adjacent to the stir jar system, as this minimizes the time needed to transfer floc samples to the LabSFLOC-2 settling chamber and any potential disruption during the subsequent floc settling process.

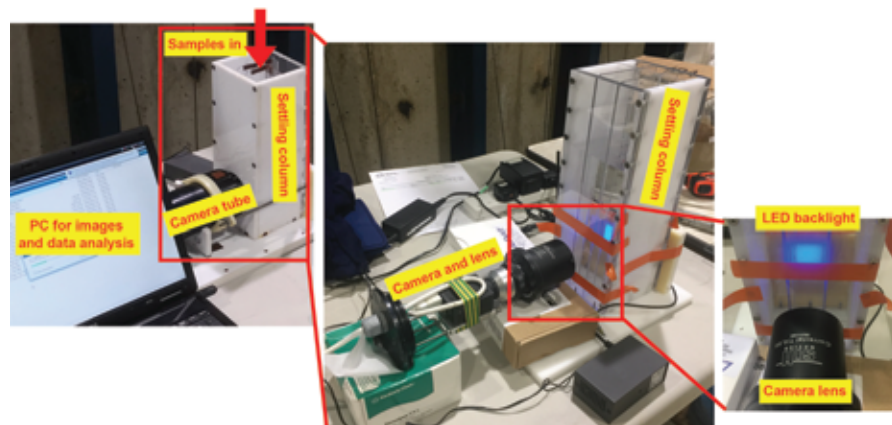
The camera views through an aperture in the settling column wall at a depth of 230 mm below the column water surface. It records all settling flocs/particles in the center of the column, which pass within a 1-mm focal depth of field, 45 mm (focal length) from the camera lens. The total image size is nominally 6 mm high and 8 mm wide. During sampling, a pipette is filled to produce a fluid head of 50 mm, which results in

a video image control sample volume nominally of 400 mm³ (1-mm image depth and 6-mm nominal video image width, with a nominal 50-mm high suspension extracted with a modified pipette). This control volume permits the LabSFLOC-2 calculated total floc mass to be accurately mass-balanced with the nominal suspended particulate matter concentration utilized in the jar test under examination. The LabSFLOC-2 camera can view particles as small as 5 μ m and as large as 8 mm. Settling velocities ranging from 0.01 to 45 mm·s⁻¹ can be measured by the LabSFLOC-2, and the system can operate within floc suspended particulate matter concentrations of a few milligrams per liter, with a practical upper operating limit of ~200 g·l⁻¹.

Settling flocs are viewed as silhouettes (to reduce image smearing) resulting from a 43 \times 35 mm, homogeneous blue (470 nm), back-illumination LED panel located at the rear of the settling column. The digital floc images are captured as non-Codec compressed AVI files at a frame rate of 7.5 Hz (one frame is 0.04 s), at a resolution of 1,600 \times 1,200 pixels, with an individual pixel nominally representing 5 μ m (confirmed by independent

FIGURE 2

Sample from an oil-bentonite case. The left plot shows the floc size and settling velocity scatters of each calculated floc. The three diagonal lines present contours of Stokes settling velocity calculated with a constant effective density (i.e., floc bulk density minus water density): pink = $1,600 \text{ kg}\cdot\text{m}^{-3}$ (equivalent to a quartz particle), green = $160 \text{ kg}\cdot\text{m}^{-3}$, and red = $16 \text{ kg}\cdot\text{m}^{-3}$. The right image is the generated OMAs as seen by the digital microscope camera (approximately $\times 40$).



calibration), connected and streamed to a laptop PC, and recorded on the internal hard drive.

The present system not only produces visible floc individual images (e.g., Figure 2, right) but also reveals all other essential quantitative floc properties. The uncompressed images are then analyzed with MATLAB software routines. During postprocessing, the HR Wallingford Ltd. DigiFloc software version 1.0 (Benson & Manning, 2013) and JavaScript can be used to semiautomatically process the digital recording image stack to obtain floc size and settling velocity spectra (e.g., Figure 2, left for oil-bentonite flocs). A modified version of Stokes' law (Stokes, 1851) permits an accurate estimate of individual floc effective density (Manning et al., 2013), which can then be utilized to calculate floc mass. In the oil-bentonite sample, resultant floc sizes (nominally mass-balanced to a suspended particulate matter concentration of $1,000\text{-mg}\cdot\text{l}^{-1}$ bentonite and 1 ml of Texas crude oil) ranged between

Summary

In the first attempt to apply the LabSFLOC-2 system in an oil-mineral flocculation study, we have combined state-of-the-art technologies/instruments in order to expand our knowledge of oil-sediment-microbial interactions and the vertical transport of oil. The preliminary laboratory experiments demonstrate that these systems can be used to produce and characterize mass settling dynamics of OMAs. Future experiments will use different oil, sediment, and microbial characteristics and turbulence levels. Statistical data on settling dynamics provided by LabSFLOC-2 will allow for a systematic analysis of the role that each factor plays in determining the resultant settling dynamics. Moving

forward, these technologies have the potential for applications to a carefully designed test matrix in order to calibrate a given modeling framework for oil-sediment-microbial interactions.

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